Root rot of canola caused by *Leptosphaeria maculans* is widespread in southern NSW and can arise from foliar infection

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ABSTRACT

Root rot symptoms caused by *Leptosphaeria maculans* were first reported in Australian canola crops in 2001 but little is known about their importance and prevalence. In a survey of over 100 commercial canola crops in southern NSW, all paddocks had plants with root rot symptoms in 2004, whilst 86% of paddocks did in 2003. Across the two seasons surveyed, there was a strong relationship (r² = 0.63) between the mean severity of root rot and stem canker recorded within a paddock. The pathway of infection under field conditions appears to be via foliar infection, whereby the pathogen enters through the leaf or cotyledon and then travels down the plant and into the roots. Root rot symptoms appear at early flowering and increase in incidence and severity as the plant matures. The current recommendations for blackleg control, such as the use of the fungicides Impact® and Jockey®, reduce the severity of root rot.

Key words: Blackleg, phoma, *Brassica napus*, Australia.

INTRODUCTION

Blackleg, caused by the fungal pathogen *Leptosphaeria maculans*, is the most devastating disease of canola in Australia and worldwide. Ascospores released from infected stubble germinate and invade cotyledons and leaves causing lesions. The fungus then grows biotrophically through the petiole to the stem base where it forms crown cankers that may girdle the stem base causing yield loss and, in severe cases, plant death. Leaf lesions and crown cankers are characteristic symptoms of blackleg.

Sosnowski et al. (2001) reported the presence of necrotic tissue in the roots of canola plants that had senesced prematurely and had no external symptoms of stem canker. *Leptosphaeria maculans* was consistently isolated from these samples and symptoms were reproduced in plants that were either inoculated on the hypocotyl or had severed roots dipped into spore suspension.

The aim of this study was to obtain a better understanding of the canola root rot caused by *L. maculans*. A survey was conducted to determine its prevalence in commercial crops. Field and glasshouse experiments were carried out to investigate the progression of the disease during crop growth, the pathway of root infection and methods for disease control.

MATERIALS AND METHODS

Prevalence of root rot symptoms in southern New South Wales

During 2003 and 2004, over 100 commercial canola crops were sampled in southern New South Wales prior to windrowing to determine the incidence and severity of root rot symptoms. Stem canker symptoms were also assessed to investigate the relationship between stem and root symptoms. Between 20 and 60 plants were randomly sampled from five locations at least 20 m apart in each paddock. Whole plants were removed from the ground and assessed for the incidence and severity of internal infection at the base of the stem and in the roots. Stems were assessed by cutting at the crown of the plant transversely with a pair of secateurs and visually assessing the area (0, 5, 10, 20, 30, 40, 50, 60, 70, 80, 90 or 100%) of the internal stem surface that had evidence of blackening. The severed root portion of the plant was then cut longitudinally and scored on a scale of 0 (no infection) to 5 (severely infected) based on the area of the cut surface that was blackened and how far the symptoms extended along the
Progression of disease symptoms during crop development

The expression of root rot symptoms was monitored in a field experiment established at Galong in southern NSW (mean annual rainfall 650 mm) in 2004. Untreated seed of cv. Grace (ABR 6.5) was sown in three replicate plots (8 m x 2 m) on June 8 into wheat stubble. Plants were assessed for leaf lesion incidence at the cotyledon to one leaf stage and at the 2 to 4 leaf stage by counting the number of plants with one or more leaf lesions out of 20 consecutive plants in each plot. Plots were sampled for root rot and stem canker incidence and severity six times between stem elongation and plant maturity. A random sample of 20 plants from each plot at each time of assessment was pulled from the ground and scored for blackleg disease in the crown and root as described above.

Pathway of root infection under field conditions

A field experiment using plots where the soil had been fumigated to a depth of 25 cm was set up at two sites in southern NSW; Wallendbeen (650 mm annual rainfall) and Ardlethan (450 mm annual rainfall) to determine the pathway of root infection under conditions of natural inoculation. Methyl bromide was injected into 8 m x 2 m plots and then covered with plastic for three days. After removal of the plastic, plots remained undisturbed for at least two weeks before sowing untreated seed of cv. Rainbow (ABR 6.5). The trial was a random block design with three blocks containing randomised combinations of soil treatment (fumigated or untreated). Fifty seedlings from each plot at Wallendbeen were assessed for the incidence of blackleg leaf lesions at the cotyledon to one leaf stage. At the 2-4 leaf stage, leaf area and aboveground biomass were measured for 12 plants taken at random from each plot due to obvious differences in seedling development between the fumigated and control plots. At plant maturity, 20 individual plants from each plot at Wallendbeen and Ardlethan were assessed for the incidence and severity of internal infection in the roots as described above.

Control of blackleg root rot with fungicides

Field experiments were conducted at Galong in New South Wales to determine the effectiveness of the current fungicide treatments for control of the root rot. In 2003, Impact® (active ingredient flutriafol) applied to the starting fertiliser at a rate of 400 ml/ha and Jockey® (active ingredient fluquinconazole) applied as a seed dressing at 2 L/tonne of seed were used both separately and in combination. In 2004, the combination treatment was not used due to some reports of yield depression. Cultivar Rainbow (ABR 6.5) was used in 2003 and cv. Grace (ABR 6.5) was used in 2004. The experiments were arranged in a randomised block design with three blocks. Each plot measured 8 m x 1.44 m. Just prior to windrowing, 20 plants from each plot were assessed for root rot and stem canker severity as described above.

RESULTS

Prevalence of root rot symptoms in southern New South Wales

In 2003, 86% of paddocks surveyed had plants with root rot, whilst in 2004 all paddocks did. Although the root rot symptom was present in 42% of plants/paddock in 2003 and 49% in 2004, the average severity of infection (0 to 5 scale) was low in both 2003 (0.85) and 2004 (1.1). Despite this low average severity of root rot, some paddocks had scores over 2.5 with over 80% of plants infected.

There was a significant linear association between the mean severity of root rot and stem infection in the paddocks surveyed ($r^2 = 0.63$) (Figure 1), whereby the severity of root rot increased with an increase in the severity of blackleg stem infection. The relationship between root and stem infection was not as strong ($r^2 = 0.41$) when the data from 4000 individual plants assessed were analysed.
Progression of disease symptoms during crop development

The incidence of plants with one or more leaf lesions increased from 60% at the cotyledon to one leaf stage to 88% at the 2 to 4 leaf stage. Symptoms of the root rot began to appear at early flowering. The incidence and severity increased slowly through the flowering and early pod filling stages but increased dramatically during late pod fill. The development of stem canker during plant development followed a similar pattern with disease symptoms beginning to appear at early flowering and increasing in incidence and severity as the plant matured (data not shown).

Pathway of root infection under field conditions

The incidence of seedling leaf lesions was similar between plants in fumigated (69%) and untreated (73%) plots at Wallendbeen. Plants grew more slowly in the fumigated plots than in the untreated plots, probably due to ammonia toxicity caused by the fumigation. Plant biomass and leaf area was 35% and 49% lower, respectively, at the 2 to 4 leaf stage in the fumigated compared to the untreated plots at Wallendbeen. At plant maturity, there was no difference in the incidence of plants with root rot in the fumigated and untreated plots at either site, however, root rot was more severe in the fumigated plots compared to the untreated plots at both sites (Table 1). The severity and incidence of root rot was higher at Wallendbeen than Ardlethan.

Table 1 Severity of root rot on a 0 to 5 scale in field experiments in NSW where soil had been fumigated with methyl bromide.

<table>
<thead>
<tr>
<th></th>
<th>Fumigated*</th>
<th>Untreated</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wallendbeen</td>
<td>3.3a</td>
<td>2.4b</td>
</tr>
<tr>
<td>Ardlethan</td>
<td>2.0a</td>
<td>1.5b</td>
</tr>
</tbody>
</table>

*Values followed by the same letter across a row are not significantly different, P=0.05

Control of blackleg root rot with fungicides

The severity of blackleg root rot was reduced with the application of the fungicides Impact® and Jockey® (Figure 2). In 2003, Impact® reduced the severity of root rot by 26% whilst Jockey had no effect. No further reduction in disease severity was observed with Impact® and Jockey® applied in combination. A 40% reduction in stem canker severity was achieved with Impact®, but again, no further reduction was gained by the addition of Jockey®. In 2004, Jockey® and Impact® reduced root rot severity by 22% and 49%, respectively. However, only Impact®
reduced the severity of stem canker. While root rot was more severe in 2003 than in 2004, stem canker severity was the same in untreated plants in both years.

Figure 2  Effect of fungicides on the severity of blackleg root rot (■) and stem canker (□) prior to windrowing at Galong, NSW, in 2003 and 2004. Treatments indicated by an asterisk (*) are significantly different from the untreated control.

**DISCUSSION AND CONCLUSION**

The presence of root rot symptoms in the majority of commercial crops surveyed in New South Wales suggests that the root rot is not a new symptom in the infection of canola by *L. maculans* but has probably not been reported previously. A number of paddocks that had never grown canola were assessed in the survey and all had plants with root rot symptoms, indicating that the pathway of *L. maculans* infection is via windborne ascospores. This is supported by the presence of root rot symptoms in plants grown in field plots in which soil organisms were killed by fumigation, thereby eliminating the possibility of infection from soilborne inoculum. A detailed study of the path of infection through the stem and into the roots is currently underway.

There was a positive relationship between the severity of root rot and stem canker in individual plants collected in the survey, however, the relationship between the mean disease severity recorded in each paddock was stronger than that for individual plants. The onset of root rot symptoms at early flowering, slow increase in severity during the flowering period and then rapid increase during senescence was similar to the pattern observed for stem canker symptoms. Although this suggests that the onset of both symptoms are triggered by the same conditions, it is interesting to note that stem canker severity was the same in both years while root rot was more severe in 2003 than in 2004 in untreated plots. Different cultivars with the same ABR were used in both years, however there may be genetic differences between cultivars in their susceptibility to the root rot form of blackleg.

Fungicides were effective in reducing the severity of blackleg root rot. Impact® was effective in both years whilst Jockey reduced the severity of infection only in 2004, although not as effectively as Impact®. In 2004, Jockey reduced the severity of root rot but did not significantly reduce the severity of stem canker. Both of these fungicides act to inhibit invasion of the leaf, thereby preventing the fungus from establishing within the plant. Since the root rot results from infection through the leaf, it is expected that a reduction in leaf penetration would
also reduce the severity of stem canker and root rot by reducing the subsequent amount of hyphae entering stem and root tissue.

Together, this work suggests that the root rot symptom is an extension of the disease caused by leaf infection and that similar measures to those recommended for stem canker are appropriate for its control. The factors that influence the progression of the disease into the root in individual plants is uncertain, but experiments are underway to determine the mechanisms and pathways involved.

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REFERENCES